CLAIMS

1. A fluorescent indicator formed by binding fluorescent molecular components having substantially identical fluorescent properties to the N- and C-terminal sides of a target sequence, to which an analytical substance binds or reacts, so as to change the three-dimensional structure of the indicator.

2. A fluorescent indicator, which comprises:

a target sequence, to which an analytical substance binds or reacts, so as to change the three-dimensional structure of the indicator;

a donor fluorescent molecular component that covalently fuses to the target sequence; and

an acceptor fluorescent molecular component that covalently fuses to the target sequence,

wherein the donor fluorescent molecule and the acceptor fluorescent molecule have substantially identical fluorescent properties, and wherein the three-dimensional structure of the target sequence is changed due to the analytical substance binding to the target sequence, and the relative positions or orientation of the donor and the acceptor molecular component are then changed, and it is highly likely that the polarization properties of fluorescence observed when such a fluorescent molecule is excited by irradiation light having certain polarization properties differ from those of the irradiation light (depolarization).

3. The fluorescent indicator according to claim 1 or 2, wherein the fluorescence molecular component is a fluorescent protein or a mutant thereof.

4. The fluorescent indicator according to any of claims 1 to 3, wherein the fluorescence molecular component is a yellow fluorescent protein or a mutant thereof.

5. The fluorescent indicator according to any of claims 1 to 4, wherein the fluorescence molecular component is a fluorescent protein Venus.

6. The fluorescent indicator according to any of claims 1 to 5, wherein the fluorescent indicator further comprises a target peptide component and a linker component, wherein the target sequence of the analytical substance further comprises a peptide-binding domain for allowing the target peptide component to bind thereto,

wherein the linker component allows the target sequence of the analytical substance to covalently fuse to the target peptide component, and the target sequence and the target peptide component covalently fuse to either the acceptor fluorescent molecular component or the donor fluorescent molecular component, and

wherein the analytical substance binding to the target sequence induces a change in the relative positions or directions of the target peptide component and the peptide-binding domain, and the relative positions or directions of the donor and acceptor molecular component are then changed, and it is thereby highly likely that the polarization properties of fluorescence observed when such a fluorescent molecule is excited by irradiation light having certain polarization properties differ from those of the irradiation light (depolarization).

7. The fluorescent indicator according to any of claims 1 to 6, wherein the target sequence is calmodulin, cGMP-dependent protein kinase, a steroid hormone receptor, a ligand-binding domain of a steroid hormone receptor, protein kinase C, inositol-1,4,5-triphosphate receptor, or recobelin.

8. The fluorescent indicator according to claim 7, wherein the target sequence of the analytic substance is calmodulin.

9. The fluorescent indicator according to claim 6, wherein the target peptide component is skeletal muscle myosin light chain kinase (skMLCKp), smooth muscle myosin light chain kinase (smMLCK), calmodulin kinase II (CaMKII), caldesmon, calspermine, phosphofructokinase, calcineurin, phosphorylase kinase, Ca^{2+}-ATPase, 59 Kda phosphodiesterase (PDE), 60 Kda phosphodiesterase (PDE), nitric oxide synthase, type I adenylyl cyclase, *Bordetella pertussis* adenylyl cyclase, neuromodulin, spectrin,

myristoylated alanine-rich C kinase substrate (MARCKS), MacMARCKS(F52), b-Adducin, heat shock protein HSP90a, human immunodeficiency virus envelope glycoprotein 160 (HIV-1 gp160), brush-boarder myosin heavy chain-I (BBMHBI), dilute myosin heavy chain (MHC), mastoparan, melittin, glucagon, secretin, vasoactive intestinal peptide (VIP), gastrin inhibitory peptide (GIP), or a calmodulin-binding domain of calmodulin-binding peptide-2 (Model peptide CBP2).

10. The fluorescent indicator according to claim 6, wherein the linker component is a peptide component.

11. The fluorescent indicator according to claim 10, wherein the linker component consists of 1 to 30 amino acid residues.

12. The fluorescent indicator according to any of claims 1 to 5, wherein the target sequence, on which an analytical substance reacts, so as to change the three-dimensional structure of the indicator, is an amino acid sequence that is cleaved with enzymes.

13. The fluorescent indicator according to any of claims 1 to 12, which is a single polypeptide.

14. The fluorescent indicator according to any of claims 1 to 13, which further comprises a localized sequence.

15. The fluorescent indicator according to any of claims 1 to 14, wherein the localized sequence is a nucleus-localized sequence, an endoplasmic reticulum-localized sequence, a peroxisome-localized sequence, a mitochondrion-localized sequence, a Goldi apparatus-localized sequence, or a cell membrane-localized sequence.

16. A method for detecting or measuring an analytical substance in a sample, which comprises:

(1) a step of allowing a sample to interact with the fluorescent indicator of any of claims 1 to 15;

(2) a step of exciting a donor component; and

(3) a step of measuring the level of fluorescence resonance energy transfer in the

indicator that reflects the concentration or activity of the analytical substance in the sample.

17. The method according to claims 16 wherein the level of fluorescence resonance energy transfer in the indicator is measured by depolarization.

18. The method according to claims 17 wherein the depolarization is measured by obtaining fluorescence anisotropy.

19. The method according to any of claims 16 to 18 wherein the sample is a living cell, and the step comprises incorporation of the fluorescent indicator into the cell.

20. The method according to claims 19 wherein the step of incorporating the fluorescent indicator into a cell comprises transfection of the cell with an expression vector containing an expression regulatory sequence that is functionally ligated to a nucleic acid sequence encoding the expression of the fluorescent indicator.

21. A nucleic acid encoding the fluorescent indicator of any of claims 1 to 15.

22. An expression vector containing the nucleic acid of claim 21.

23. A transformant having the nucleic acid of claim 21 or the expression vector of claim 22.